



QUALITY CONTROL PROCEDURES

I INTRODUCTION

Nutrient Gelatin is a medium for determining the ability of relatively nonfastidious microorganisms to liquefy gelatin as an aid to their identification.

II PERFORMANCE TEST PROCEDURE

1. Inoculate tubes with an inoculating needle by stabbing half the depth of the medium using 18- to 24-h **Trypticase™** Soy Agar with 5% Sheep Blood (TSA II) cultures of the organisms listed below.
2. Incubate tubes with loosened caps at 35 ± 2°C in an aerobic atmosphere.
3. Examine the tubes for up to 14 days for growth and gelatin liquefaction. Use an uninoculated control tube for comparison. At each interval, transfer the tubes to a refrigerator or ice bath for a sufficient time period to determine whether liquefaction has or has not occurred. It is important that the tubes not be shaken during the transfer from incubator to refrigerator. When reading results, invert the chilled tubes to test for solidification or liquefaction.
4. Expected Results

Organisms	ATCC™	Recovery and Reaction
* <i>Proteus vulgaris</i>	8427	Growth with liquefaction
* <i>Serratia liquefaciens</i>	27592	Growth with liquefaction
* <i>Escherichia coli</i>	25922	Growth without liquefaction
<i>Staphylococcus aureus</i>	25923	Growth with liquefaction

*Recommended organism strain for User Quality Control.

III ADDITIONAL QUALITY CONTROL

1. Examine tubes as described under "Product Deterioration."
2. Visually examine representative tubes to assure that any existing physical defects will not interfere with use.
3. Incubate uninoculated representative tubes at 20–25°C and 30–35°C and examine after 5 days for microbial contamination.

PRODUCT INFORMATION

IV INTENDED USE

Nutrient Gelatin is used for the detection of gelatin liquefaction by microbial species.

V SUMMARY AND EXPLANATION

Nutrient Gelatin is made in accordance with the formula formerly used in the examination of water, sewage, and other materials of sanitary importance.¹ Gelatin liquefaction is one of the characteristics used in the classification of members of the *Enterobacteriaceae* and nonfermenting gram-negative bacteria. The use of Nutrient Gelatin for determining gelatin liquefaction patterns is considered to be the "standard" method for taxonomic studies, since the rate of liquefaction is important in the characterization of groups within the *Enterobacteriaceae* family as well as other groups of microorganisms.^{2,3} Edwards and Ewing consider gelatin liquefaction to be an essential test for differentiation of enteric bacilli.⁴

Nutrient Gelatin is used chiefly for identification of pure cultures of bacteria which are not particularly fastidious in regard to nutritional requirements.

VI PRINCIPLES OF THE PROCEDURE

The peptone and beef extract supply sufficient nutrients for the growth of nonfastidious bacterial species. The gelatin is the substrate for the determination of the ability of an organism to produce gelatinases, which are proteolytic-like enzymes active in the liquefaction of gelatin.

VII REAGENTS

Nutrient Gelatin

Approximate Formula* Per Liter Purified Water

Peptic Digest of Gelatin	5.0 g
Beef Extract	3.0 g
Gelatin	120.0 g

*Adjusted and/or supplemented as required to meet performance criteria.

Warnings and Precautions: For *in vitro* Diagnostic Use.

Tubes with tight caps should be opened carefully to avoid injury due to breakage of glass.

Observe aseptic techniques and established precautions against microbiological hazards throughout all procedures. After use, prepared tubes, specimen containers and other contaminated materials must be sterilized by autoclaving before discarding.

Storage Instructions: On receipt, store tubes in the dark at 2–25°C. Avoid freezing and overheating. Do not open until ready to use. Minimize exposure to light. Tubed media stored as labeled until just prior to use may be inoculated up to the expiration date and incubated for the recommended incubation times. Allow the medium to warm to room temperature before inoculation.

Product Deterioration: Do not use tubes if they show evidence of microbial contamination, discoloration, drying or other signs of deterioration.

VIII SPECIMEN COLLECTION AND HANDLING

This product is not intended for use directly with specimens or mixed cultures. The organism to be tested must first be in pure culture.

IX PROCEDURE

Material Provided: Nutrient Gelatin

Materials Required But Not Provided: Ancillary culture media, reagents, quality control organisms and laboratory equipment as required.

Test Procedure: Observe aseptic techniques.

Using a heavy inoculum (growth from an 18–24 h pure culture), stab the tubes of Nutrient Gelatin with an inoculating needle directly down the center of the medium to a depth of approximately one half inch from the bottom of the tube. Incubate tubes, including an uninoculated control, aerobically at $35 \pm 2^\circ\text{C}$ for 24–48 h and up to 14 days.

User Quality Control: See "Quality Control Procedures."

Quality Control requirements must be performed in accordance with applicable local, state and/or federal regulations or accreditation requirements and your laboratory's standard Quality Control procedures. It is recommended that the user refer to pertinent CLSI (formerly NCCLS) guidance and CLIA regulations for appropriate Quality Control practices.

X RESULTS

At various intervals during the incubation process, examine the tubes for growth (turbidity) and liquefaction. Use uninoculated control tubes for comparison. At each interval, tighten caps and transfer the tubes to a refrigerator or ice bath for a sufficient time period to determine whether liquefaction has or has not occurred. It is important that the tubes not be shaken during the transfer from incubator to refrigerator. When reading results, invert the chilled tubes to test for solidification or liquefaction.³ Consult appropriate texts for results with specific organisms.^{3-5,7}

XI LIMITATION OF THE PROCEDURE

For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification. Consult appropriate texts for detailed information and recommended procedures.⁵⁻¹⁰

XII AVAILABILITY

Cat. No.	Description
220974	BBL™ Nutrient Gelatin Deeps, 8 mL, Pkg. of 10 size K tubes

XIII REFERENCES

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5. Murray, P.R., E.J. Baron, J.H. Jorgensen, M.A. Pfaller, and R. H. Tenover (ed.). 2003. Manual of clinical microbiology, 8th ed. American Society for Microbiology, Washington, D.C.
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7. Holt, J.G., N.R. Krieg, P.H.A. Sneath, J.T. Staley, and S.T. Williams (ed.). 1994. Bergey's Manual™ of determinative bacteriology, 9th ed. Williams & Wilkins, Baltimore.
8. MacFaddin, J.F. 2000. Biochemical tests for identification of medical bacteria, 3rd ed. Lippincott Williams & Wilkins, Baltimore.
9. Koneman, E.W., S.D. Allen, W.M. Janda, P.C. Schreckenberger, and W.C. Winn, Jr. 1997. Color atlas and textbook of diagnostic microbiology, 5th ed. Lippincott-Raven, Philadelphia.
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